## Sampling and Testing Recommendations for the Detection of Cry9C Protein in Hybrid Seed Corn

Collect a representative sample from each seed lot using the recommended sampling protocols established by the Grain Inspection, Packers, and Stockyards Administration (GIPSA) of USDA or an equivalent method. Grain sampling methods prescribed by GIPSA include methods for sampling moving grain streams and static grain lots. The diverter type (DT) sampler is the most common sampling device for sampling from a grain stream. The DT traverses a moving grain stream and, per specific timer settings, diverts a small slice of the grain stream which is collected as a subsample. The subsamples are combined to obtain a sample for the lot.

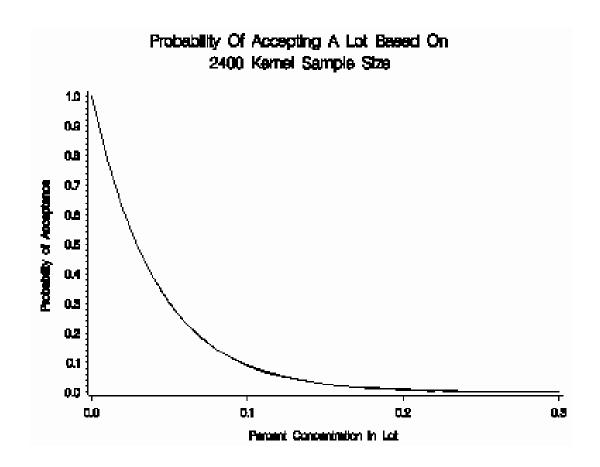
Systematically subdivide and reduce the collected sample to a 2400 kernel sample. Analyze all 2400 kernels for the presence of Cry9C protein. The operating characteristic curves and table for a sample plan of 2400 kernels are provided in the appendix. For example, a seed lot containing 0.19% StarLink corn would be rejected 99% of the time using the 2400 kernel sample size.

This sample plan assumes that one biotech kernel in the 2400 kernel sample can be detected by the analytical method used. In practice, however, analytical methods may have limits on the concentrations that are detectable. For example, an analytical method may only be able to reliably detect one biotech kernel in a sample of 400 kernels. Consequently, to use this methodology the 2400 kernel sample must be thoroughly mixed and divided into subsamples of 400 kernels or less and then each subsample ground and tested.

GIPSA has conducted studies to validate the TraitCheck Bt9 Lateral Flow Test (Strategic Diagnostics Inc.) and the Cry9C QuickStix<sup>TM</sup> test (EnviroLogix Inc.) using yellow corn. GIPSA verified the test kits are capable of detecting the presence of the Cry9C protein at the minimum detectable level of 1 StarLink kernel in 400 corn kernels (0.25%) for the TraitCheck Bt9 and 1 StarLink kernel in 500 corn kernels (0.2%) for the Cry9C QuickStix<sup>TM</sup>, as claimed by the manufacturers. Other testing methods are commercially available providing a wide range of detection capability. Caution must be taken to ensure that the sample size analyzed is consistent with the detection capability of the methodology used.

The person performing the analysis should be adequately trained. Seed company staff may find the following GIPSA information useful when establishing company operating procedures. Contact GIPSA at 202-720-0252 or <a href="www.usda.gov/gipsa">www.usda.gov/gipsa</a>.

- GIPSA Directive 9181.1 Testing for StarLink<sup>TM</sup> Corn
- Grain Inspection Handbook, Book 1, Grain Sampling
- Sampling for the Detection of Biotech Grains



Probability (%) of accepting a corn lot at various levels of concentration (% of corn kernels containing Cry9C protein) based on a 2400 kernel sample size.

Concentration	Probability of Acceptance	Concentration	Probability of Acceptance	Concentration	Probability of Acceptance
0	100	0.10	9.1	0.20	0.8
0.01	78.7	0.11	7.1	0.21	0.6
0.02	61.9	0.12	5.6	0.22	0.5
0.03	48.7	0.13	4.4	0.23	0.4
0.04	38.3	0.14	3.5	0.24	0.3
0.05	30.1	0.15	2.7	0.25	0.2

0.06	23.7	0.16	2.1	0.26	0.2
0.07	18.6	0.17	1.7	0.27	0.2
0.08	14.6	0.18	1.3	0.28	0.1
0.09	11.5	0.19	1.0	0.29	0.1
				0.30	0.1